Illinois River Watershed
Soil and Litter/Manure Sampling Protocol

I. Selection of Sampling Locations

A. Sample locations will be selected from either contract growers farms or company-owned facilities for up to seven Integrators. Between one and three farms/facilities will be selected from each Integrator. At each of these farms/facilities, litter from the poultry houses will be collected. Collection of litter from the poultry houses will be conducted by the owner or his representative. Fields where documentation of litter application from a specific farm and Integrator is available from the Oklahoma Department of Agriculture, Food and Forestry will also be selected for sample collection. Between one and three fields will be selected from each Integrator. Field locations selected will be within the Illinois River Watershed (see Exhibit "A", a map showing the boundary of the Illinois River Watershed, roads, towns and public land survey grid). Personnel from the Oklahoma Department of Agriculture, Food and Forestry will collect the samples from the fields. If permission is obtained from the owner of the field, staff from CDM may also entry the field and assist in sampling. Neither personnel from the Department of Agriculture, Food and Forestry or CDM will sample litter from poultry houses. Considerations for sample collection from farms/facilities and fields include:

1. Poultry litter/manure has been consistently produced,
2. Poultry litter/manure is currently being generated,
3. Poultry litter/manure has been consistently (every year for the at least the past 3 years) applied to land (Litter Application Locations, "LALs") associated with the Farm/Facility,
4. Availability of land upon which poultry litter/manure has not been applied (Control Locations, "CLs").

B. The locations should contain the following information for each associated Farm/Facility:

1. Name of Farm/Facility owner and Farm/Facility contact person,
2. Physical address and location (section-township-range) of Facility,
3. Contact address of Farm/Facility owner or Farm/Facility contact person,
4. Contact phone number of Farm/Facility owner or Farm/Facility contact person,

5. Whether or not one or more LALs can be accessed at the Farm/Facility,
6. The physical location of each LAL.
7. Whether or not one or more CLs can be accessed at the Farm/Facility.
8. The physical location of each CL.
9. Whether or not a litter/manure and/or nutrient management plan has been prepared for LALs at the Farm/Facility,
10. Estimates of the amounts, rates and dates of prior litter/manure applications to each LAL at the farm/facility,
11. Estimates of litter treatment or amendments added to each LAL (e.g., alum, etc), if any, and information as to amount, rate and dates of application
12. Number, type, physical size and capacity of poultry grow houses (or other poultry/egg production facilities, as appropriate) operated at Farm/Facility ("Poultry Houses").

C. Between one and three Farms/Facilities from each Integrator will be selected for soil and litter/manure sampling. Either facilities containing turkeys or broilers will be selected. One facility with pullets and one facility with layers will also be selected. Currently 8 facilities have been identified as the locations for sampling. Additional backup facilities have also been identified from each Integrator. Each of these Facilities will have associated LALs, but may or may not have associated CLs.

D. If available, at least three Farms/Facilities with CLs will be selected.

E. If available, LALs and CLs associated with a single Farm/Facility be sampled.

F. If only the CL at a Farm/Facility is sampled, the Poultry Houses at that facility will not be sampled.

G. The total number of LALs selected will not exceed twenty-four (24).

H. The total number of CLs selected will not exceed eight (8).

II. Sampling Documentation

A. Sampling Log Book and Sampling Forms

1. A Sampling Log Book and Sampling Forms shall be maintained.
2. Example forms are provided in Exhibit B. Pages in the Sampling Log Book will reference specific Sampling Forms by use of the Facility Identification.

3. The Sampling Log Book shall be bound and shall be constructed of waterproof paper.
4. Entries in the Sampling Log Book or the on the Sampling Form shall be made in black permanent ink.
5. Each page of the Sampling Log Book shall be dated.
6. The preparer shall initial each page of the Sampling Log book.
7. For each Farm/Facility sampled, the following information shall be recorded in the Sampling Log Book or on the Sampling Forms:
    a. Name, address and phone number of the Farm/Facility owner,
    b. Identification of the Farm/Facility, FAC1 – FAC8,
    c. Name, address and phone number of the Farm/Facility operator,
    d. Name, address and phone number of the Integrator responsible for the Farm/Facility,
    e. Names, addresses and phone numbers of persons who have spread litter/manure on LALs associated with the Farm/Facility,
    f. The amounts, rates and dates of prior litter/manure applications to specific LALs at the Farm/Facility (confirm State Reports),
    g. The existence of prior soil sampling data for LALs or CLs at the facility (yes or no),
    h. The water supply for the Farm/Facility,
    i. The legal description (qtr-qtr-qtr-sec-twp-rng) of the property related to the Farm/Facility,
    j. The legal description (qtr-qtr-qtr-sec-twp-rng) of the CLs at the Farm/Facility,
    k. Type of animals generating litter (broilers, layers, pullets, turkeys, etc.),
    l. Number of flocks of birds that have used the litter that is sampled,
    m. The number of birds in each flock that have used the litter that is sampled,
    n. The time since birds last used the litter,

o. Litter treatment (e.g. alum amendment), if any, and information as to amount, rate and date or dates of treatment,

p. Information as to any other fertilizers, chemicals or soil amendments added during the last five years,

q. Use of each LAL by cattle (yes or no) and typical number of cattle.

r. Specific information listed within this protocol,

s. Sketch map of each LAL with approximate dimensions; indicate local features on the sketch (vegetation, water bodies, adjacent fields, location of poultry houses, roads, old fence rows, livestock feeding areas, livestock grazing areas, etc); dimensions and features can also be placed on the aerial photographs,

t. Land slope of each LAL (or LAL sub-area),

u. Distance to nearest water body,

v. Notes on weather (temperature, wind, last precipitation event, etc),

w. Type of vegetation currently on the LAL, if any, and any known vegetation grown in past 5 years,

x. Use of adjacent fields, and;

y. Other information as appropriate or relevant.

B. Photographic Record

1. A photographic record shall be made and maintained for all sampling activities on the LAL. Pictures of the LALs, CLs and the outsides of the poultry house will be taken. No pictures of sampling activities inside the poultry houses will be taken.

2. All photographs made shall be time and date stamped.

C. Chain-of-Custody

1. A Chain-of-Custody shall be prepared for each set of samples transferred to the soil and litter processing lab (CDM Support Laboratory in Denver, Colorado). A second chain-of-custody will be prepared at the processing lab for the analytical laboratory.

2. The Chain-of-Custody to the soil processing lab shall, at a minimum, contain the following information:

a. The project name, *Illinois River Watershed Soil and Litter/Manure Sampling,*

b. Name of person or entity collecting samples,

c. Signature blocks with dates and times for all persons having custody (sampler, shipper, processing laboratory, etc),

d. For each sample related to a Chain-of-Custody:

i. The unique numeric identifier on the submitted sample container/bag,

ii. The date and time the sample was collected,

iii. The sample "matrix" (i.e. SOIL or LITTER), and;

III. Soil Sampling

A. Litter Application Locations (LALs) and Control Locations (CLs)

1. Permissible Soil and Weather Conditions

a. Soils cannot be sampled if water saturated.

b. Soils cannot be sampled during precipitation events.

2. Division into Sampling Areas

a. A Sampling Area is an area within a LAL or CL that is reasonably homogenous with respect to soil types, soil properties, topography, landscapes, management history (to the extent known), and other relevant factors, as appropriate.

b. For each LAL or CL sampled, the LAL or CL shall be divided into a maximum of four Sampling Areas, identified as A, B, C and D.

c. Sampling Areas identified within the LAL or CL shall be a minimum of approximately one acre and shall not exceed approximately 10 acres.

d. In making determinations concerning the division of the LAL or CL into Sampling Areas, the person or persons making those determinations shall consult the relevant USDA/NRCS soil survey and USGS topographic map and/or data. The data consulted shall be identified by reference in the Sampling Log Book.

e. The person or persons who make the determinations concerning the division of the LAL or CL into Sampling Areas shall prepare a sketch map of the LAL or CL and its constituent Sampling Areas. This sketch map shall show the approximate boundaries of each Sampling Area and the estimated area of each Sampling Area. This information can also be recorded on the aerial photographs.

3. Identification of Sub-Sampling Locations

    a. A Sub-Sampling Location is an area within a Sampling Area at which individual soil samples will be collected.

    b. A total of 20 sub-sampling locations shall be sampled for each Sampling Area.

    c. Sub-Sampling Locations shall be identified by following the procedure outlined in Exhibit C, Sample Location Protocol for Each of the Sampling Areas, A thru D.

    d. The selection of Sub-Sampling Locations shall avoid:

        i. Old fence rows,

        ii. Livestock feeding areas,

        iii. Livestock loafing areas, and;

        iv. Localized conditions atypical of the Sampling Area.

    e. The geographic coordinates (Latitude and Longitude) of the first Sub-Sampling Location in each Sampling Area or a corner of the Sampling Area shall be determined using a Global Positioning System (GPS) receiver accurate to at least five (5) meters. These geographic coordinates shall be recorded in the Sampling Log Book.

    f. Representative Sub-Sampling locations shall be documented with a time and date stamped photograph.

4. Soil Samples to be collected at each Sub-Sampling Location

    a. For purposes of this Protocol, a Sub-Sampling Location shall be an area defined by a triangle with three-foot sides with the middle placed on the Sub-Sampling Location.

    b. At each Sub-Sampling Location, core samples with a length of at least six inches will be collected at the corners of the triangle. The samples will be divided into three separate soil samples as follows:

        i. Four (4) to Six (6) Inch Sample depth. This two inch sample will be collected by measuring the length of the core from the top of the sample. The two inch section of core will be placed in a plastic bag with the appropriate identification.

        ii. Two (2) to Four (4) Inch Sample depth. This two inch sample will be collected by measuring the length of the core from the top of the sample.

The two inch section of core will be placed in a plastic bag with the appropriate identification.

iii. Zero (0) to Two (2) Inch Sample depth. This two inch sample will be collected by measuring the length of the core from the top of the sample. The two inch section of core will be placed in a plastic bag with the appropriate identification.

iv. One core sample will be collected at each corner of the triangle until enough sample is collected (the bag is full, approximately 200 to 300 grams).

c. The soil samples collected at each sub-sampling location shall be collected with soil probe coring devices marked for 6-inch, 4-inch and 2-inch depths. The diameters of all soil corers used should be the same, and should be of a diameter consistent with general practice for agricultural soil sampling.

d. Whenever a soil sample is to be collected, thatch and other plant residue shall be removed from the soil surface prior to pushing the soil probe core into the soil.

i. In the event that soil conditions do not permit the use of a soil probe coring device, samples may be collected with a shovel.

ii. Thatch and other plant residue shall be removed prior to collecting a sample with a shovel.

iii. When a shovel is used for collection the following procedure shall be followed:

(A) At each sub-sampling location, dig a hole at least 6 inches deep.

(B) During excavation, material from zero to 2 inches should be placed in a bag appropriately labeled for the depth. Then the material from 2 to 4 inches should be placed in a separate bag. And finally the material from 4 to 6 inches should be placed in a separate bag.

(C) Material from each depth interval may be placed on a plastic sheet to facility sample collection.

e. Representative soil samples collected shall be documented with a time and date stamped photograph.

5. Handling of Samples

a. All individual samples from each sub-location will be placed in individual plastic bags (double bagged). The sample number will be placed between the inner and outer plastic bag. Each sampling area (up to four sampling areas per LAL) will have 60 individual samples (20 sub-sample locations x 3 sample depths = 60 samples). All samples will be shipped to the soil/litter processing laboratory for compositing.

b. Compositing of Samples will be performed at the soil processing laboratory. Procedures are provided in Exhibit D, Soil and Litter Sample Compositing Protocol.

c. Field QA/QC Samples (Soils)

i. Field Duplicate Samples will be created at the soil processing lab (see Exhibit D).

ii. Blind Standard: A blind standard of a certified reference soil shall be sent to the analytical lab for every 50 samples send to the analytical laboratory. The blind standard will be sent by the soil processing lab (see Exhibit D).

iii. Decontamination Blank: a sample of the final decontamination rinsate shall be collected and forwarded to the soil processing laboratory for analysis at a frequency of one decon rinsate collected after sampling is completed at a facility. The decon blank will be generated in the field using laboratory grade distilled water.

6. Decontamination Procedures

a. After each collection of the 20 sub-samples, all equipment that will be reused will be decontaminated. That is, decontamination will occur between every Sampling Area.

b. Decontamination will consist of removing all soil or other material by brushing/scraping the equipment. The equipment will then be washed with a phosphate free soap solution. This will be followed by a rinse with distilled water and then with 6 percent bleach.

c. All decontamination water and solutions will be contained and removed from the facility. The

decontamination fluids will be tested and disposed appropriately.

IV. Litter/Manure Sampling

A. General Conditions

1. All litter/manure samples shall be collected by the farm owner or his designated representative.
2. All litter/manure samples shall be collected with litter/manure in place within Poultry Houses.
3. Litter/manure may be sampled at any time regardless of weather conditions.
4. More than one Poultry House may be sampled at a Farm/Facility. The litter/manure from each house shall be maintained as a separate sample. Typically two to three houses will be sampled at each farm/facility.

B. Location and Distribution of Poultry House Sub-Sample Collection Points

1. Broiler or Pullet Houses
   a. A schematic of the sampling scheme for the collection of litter/manure samples is shown in Exhibit "E".
   b. Sub-samples are collected from approximately 1/3 house-width zones.
   c. Approximately six samples are collected from each zone.
   d. Sub-samples should be located so as to obtain two samples from around the waters, feeders and walls on each side of the house.
   e. Sub-samples are spaced at 20 to 25 pace intervals.
   f. Sub-sampling locations alternate between the "sides" of each zone (i.e. a "zig-zag" pattern is traversed between sampling locations within a zone).
   g. Sub-samples collected from adjacent zones should not be immediately adjacent.
2. Breeder Houses (partially slatted)
   a. Sub-samples shall be collected from both slatted and litter areas.
   b. Twenty (20) sub-samples shall be collected

c. Sub-samples shall proportionally represent the relative aerial proportion of slatted and litter areas; for example if 2/3 of the house is under slats, and 1/3 is litter area, 14 litter/manure samples should be collected form under the slats and 7 litter/manure samples should be collected from the litter area.

d. Sub-samples taken beneath slats shall be as fully penetrating of the manure as possible and shall be distributed so as to obtain a representative sample of the entire slatted area.

e. Sub-samples from litter areas shall be collected in the same manner (i.e. "zig-zag" pattern) as used for broiler or pullet houses. (See Exhibit "E").

3. Other Circumstances

a. Sampling of litter/manure within a Poultry House for circumstances and conditions other than those described for Broiler, Pullet or Breeder Houses shall be conducted so as to obtain a representative sample of the litter/manure within that Poultry House.

b. The circumstances or conditions requiring a variation from the sampling protocol described for Broiler, Pullet or Breeder Houses shall be documented in the Sampling Log Book.

c. A description of the method(s) and procedures used to collect a representative sample of the litter/manure within a Poultry House in which the sampling protocol for Broiler, Pullet or Breeder Houses cannot be followed shall be documented in the Sampling Log Book.

d. The method(s) and procedures used to collect a representative sample of the litter/manure within a Poultry House in which the sampling protocol for Broiler, Pullet or Breeder Houses cannot be followed shall follow the principles embodied in the reference materials attached as Exhibit "E".

C. Documentation

1. The geographic coordinates (Latitude and Longitude) of the approximate center of each sampled poultry house along its long axis shall be determined using a Global Positioning System (GPS) receiver accurate to at least five (5) meters. This measurement shall be made outside the poultry house. These geographic coordinates shall be recorded in the Sampling Log Book.

2. All sub-samples shall be collected with an appropriate solid manure sampling device.
3. All samples from litter areas shall be collected through the full thickness (surface to base) of the litter/manure.
4. All samples from slatted areas shall, to the extent possible, be collected through the full thickness (surface to base) of the litter/manure.
5. Immediately after collection, all sub-samples shall be placed in a plastic bag contained inside a 5-gallon plastic bucket.
6. For partially slatted houses, sub-samples from slatted and litter areas shall be composited together.
7. The 5-gallon bucket will be given to staff from the Oklahoma Department of Agriculture, Food and Forestry.

D. Sample Compositing

1. The plastic bag inside of the 5-gallon bucket will be closed with a tie. The appropriate chain-of-custody will be placed inside each bucket and the bucket sealed. The bucket will be transported to the soil/litter processing lab. No mixing or compositing will occur in the field.
2. Field QA/QC Samples (Manure/Litter)
    a. Field Duplicate Samples will be created in the soil/litter processing lab (see exhibit D).
    b. Decontamination Blank (created in the field): a sample of the final decontamination rinsate shall be collected and forwarded to the processing lab to send to the analytical lab for analysis at a frequency of one decon rinsate for every facility.
    c. Blind Standard (created at the soil/litter processing lab): A blind standard of a certified reference soil shall be sent for every 10 litter samples sent to the analytical laboratory (two blind standards).

V. Person(s) Collecting Samples and Observing Sampling

A. Personnel from the Oklahoma Department of Agriculture, Food and Forestry will conduct the soil sampling from each LAL. If permission from the owner is obtained to go on the field, personnel from CDM will also assist in sampling of the field. If permission is not obtained, CDM personnel will remain at the edge of the field to receive the samples. CDM personnel can process samples, chain-of-custody, coordinate shipping, etc.

B. Neither personnel from the Oklahoma Department of Agriculture, Food and Forestry or CDM will collect litter from the poultry houses.

VI. Identification of Samples

A. Identification of Samples

1. Identifying information to be recorded on the sample label for soil samples:

a. Alphanumeric identification of the LAL or CL: LAL1 – LAL24, CL1 – CL8. The log book will be used to record the farm and location of each LAL or CL.

b. Alphanumeric identification of the Sampling Area: A – D

c. Alphanumeric identification of the Sub-sample location: 1 - 20

d. Alphanumeric identification of the depth of collection (i.e. –2, -4, -6)

e. The following sample number is an example of the soil sample taken from LAL field number 5, sampling area B, sub-sample location 18, and a depth of 2 inches:

LAL5-B-18-2

f. For samples submitted to the analytical lab, additional alphanumeric identification of the type of sample will be added to the end of the identification number:

i. A= laboratory sample

ii. B = laboratory duplicate

iii. C = reference soil (standard)

iv. D = decontamination blank (added to field samples)

v. E = laboratory QA/QC (extra volume)

g. Date of sample collection (only on chain-of-custody),

h. Time of sample collection (only on chain-of-custody),

i. Initials of the person collecting the sample (only on chain-of-custody).

2. Identifying information to be recorded on the sample label for litter/manure samples:

a. Alphanumeric identification of the Facility: FAC1 – FAC8.

b. Alphanumeric identification of the Poultry House: A – C

c. The following sample number is an example of the litter sample taken from facility number 5 and poultry house B:

FAC5-B

d. Samples sent to the analytical laboratory will have alphanumeric identification of the type of sample added to the end of the number:

i. A= laboratory sample

ii. B = laboratory duplicate

iii. C = reference soil (standard)

iv. D = decontamination blank (added in the field)

v. E = laboratory QA/QC (extra volume)

e. Date of sample collection (only on chain-of-custody),

f. Time of sample collection (only on chain-of-custody),

g. Initials of the person collecting the sample (only on chain-of-custody).

B. When sending samples to the analytical lab, identifying information marked on the sampling container shall be protected by covering the identifying information with clear plastic tape.

VII. Shipment of Samples to the soil/litter processing laboratory and to the analytical laboratory

1. Once placed in sampling containers (plastic bags or jars), samples shall be placed on blue ice (sealed in plastic bags) within insulated protective containers.

2. If possible, samples shall be shipped immediately via overnight shipment to the analytical laboratory.

3. In no event, shall samples be held more than 24 hours before shipment.

4. Samples shall be sent to the laboratory under a Chain-of - Custody.

5. A custody seal will be place on the outside of the container across the area between the lid and the container. The custody seal will be signed.

6. The Chain-of-Custody shall be sealed in a plastic bag and placed within the insulated protective container holding those samples to which it refers.

VIII. Analytical

A. Laboratory

1. The laboratory conducting the analyses shall be experienced in conducting the specified analyses and shall have certifications to conduct the specified analyses.
2. All analyses and sample preparation shall be conducted using accepted and published protocols and/or methods.

B. Analytical Protocols

1. Soil samples will be analyzed for one of two sets of parameters (short list or long list). Table 1 provides the parameters and analytical methods for the short and Table 2 provides the parameters and analytical methods for the long list.
2. Litter samples will be analyzed for Table 2 parameters.

Table 1: Short List Parameters - Soil

Parameter	Method
Moisture content (%)	Gravimetric (105C)
Organic matter	Walkley-Black (Modified)
Soil pH	Water 1:1
Soil Conductivity	Water 1:2
Total Nitrogen	Kjeldahl, modified
Total Aluminum (Al)	EPA SW-3050/6010
Total Phosphorous (P)	EPA SW-3050/6010
Total Arsenic (As)	EPA SW-3050/6010
Total Copper (Cu)	EPA SW-3050/6010
Total Zinc (Zn)	EPA SW-3050/6010

Table 2: Long List Parameters – Manure and Soil

Parameter	Method
Moisture content (%)	Gravimetric (105C)
Organic matter	Walkley-Black (Modified)
Texture (% sand, silt and clay)	Hydrometer ASTM-D422
Soil pH	Water 1:1
Soil Conductivity	Water 1:2
Total Phosphorous (P)	EPA SW-3050/6010
Mehlich-III Phosphorous (Mehlich-III P)	Mehlich III (ICP)
Soluble Phosphorous	Water 1:10, Bull.396, pg 17
Soluble nitrate	Water 1:10
Total Nitrogen	Kjeldahl, modified
Soluble ammonium	Water 1:10
Soluble sulfate	Water 1:10
Soluble chloride	Water 1:10
TAL Metals	EPA SW-3050/6010
Total Molybdenum (Mo)	EPA SW-3050/6010
Bacteria:	
Total coliform	SM-9221B
enterococcus	SM-9221F
Fecal coliform	SM-9230B
e-coli	SM-9221F
staphylococcus	MPN
campylobacter	MPN
salmonella	MPN
17β-estradiol, estrone, estriol	LC-MS-MS

C. Anticipated number of samples

1. Soils: Assuming four areas and three depths will be collected at 24 LALs and 8 CLs, 384 soil samples (32 fields x 4 areas x 3 depths) will result (after compositing) and be analyzed. In addition, 38 duplicates (1 in 10); 8 decon rinsates (one after each facility); and 8 standards (1 in 50) will be collected. Two samples for each LAL and CL (32 x 2 = 64) will be selected for Table 2 analyses. Six of the duplicates, one of the standards and 1 of the rinsate samples will be analyzed for Table 2. The remaining samples (320 soil samples, 30 duplicates, five standards and seven rinsates) will be analyzed for Table 1 parameters. In total,

438 samples will be analyzed (362 for Table 1 and 72 for Table 2 parameters).

2. Litter: Assuming three houses from each of eight integrators, 24 litter samples will be collected. In addition, 8 duplicates, two decon rinsates and two standards will be analyzed. In total, 36 samples will be analyzed for Table 2 parameters.

D. Data Reporting

1. Data from the laboratory shall be reported in both electronic and paper reports.
2. Data reports shall include all quality control data generated, including results for duplicates, blanks and spikes, as applicable. A level 3 data quality report will be provided by the laboratory.
3. Data reports shall include a copy of the Chain of Custody accompanying each set of samples submitted

IX. Biosecurity, Decontamination of Equipment and Personal Protective Equipment

A. All persons engaged in sampling, observing sampling or documenting sampling under this protocol shall follow appropriate biosecurity precautions.

B. Soils

1. To the extent possible, disposable sampling equipment should be used.
2. All reusable sampling equipment shall be decontaminated using a non-phosphate detergent and three deionized water rinses between Sampling Areas.

C. Litter/Manure

1. To the extent possible, disposable sampling equipment should be used.
2. All reusable sampling equipment shall be decontaminated using a non-phosphate detergent and three deionized water rinses between poultry houses.

D. Health and Safety Plan: A health and safety plan that is specific to this sampling protocol will be prepared and reviewed by all samplers.

X. Exhibits

A. Map showing the boundary of the Illinois River Watershed, Roads, Towns and Public Land Survey Grid.

B. Forms for recording field information.

C. Sample Location Protocol for Each of the Sampling Areas, A thru D.

D. Soil and Litter Sample Compositing Protocol.

E. Schematic of the Sampling Scheme for the Collection of Litter/Manure Sub-Samples.

F. Reference Materials Relied Upon in Preparing this Document

1. Zhang, H. and Johnson, G. 2003. How to get a good soil sample. Oklahoma State University Cooperative Extension Service Fact Sheet F-2207. Available at http://osuextra.okstate.edu/pdfs/F-2207web.pdf

2. Zhang, H., Hamilton, D. W. and Britton, J. G. 2002. Sampling Animal Manure. Oklahoma State University Cooperative Extension Service Fact Sheet F-2248. Available at http://osuextra.okstate.edu/pdfs/F-2248web.pdf

3. Eucha/Spavinaw Watershed Management Team. Undated. Soil Sampling Protocol.

4. Eucha/Spavinaw Watershed Management Team. Undated. Steps for Pulling Litter Samples.

Exhibit A

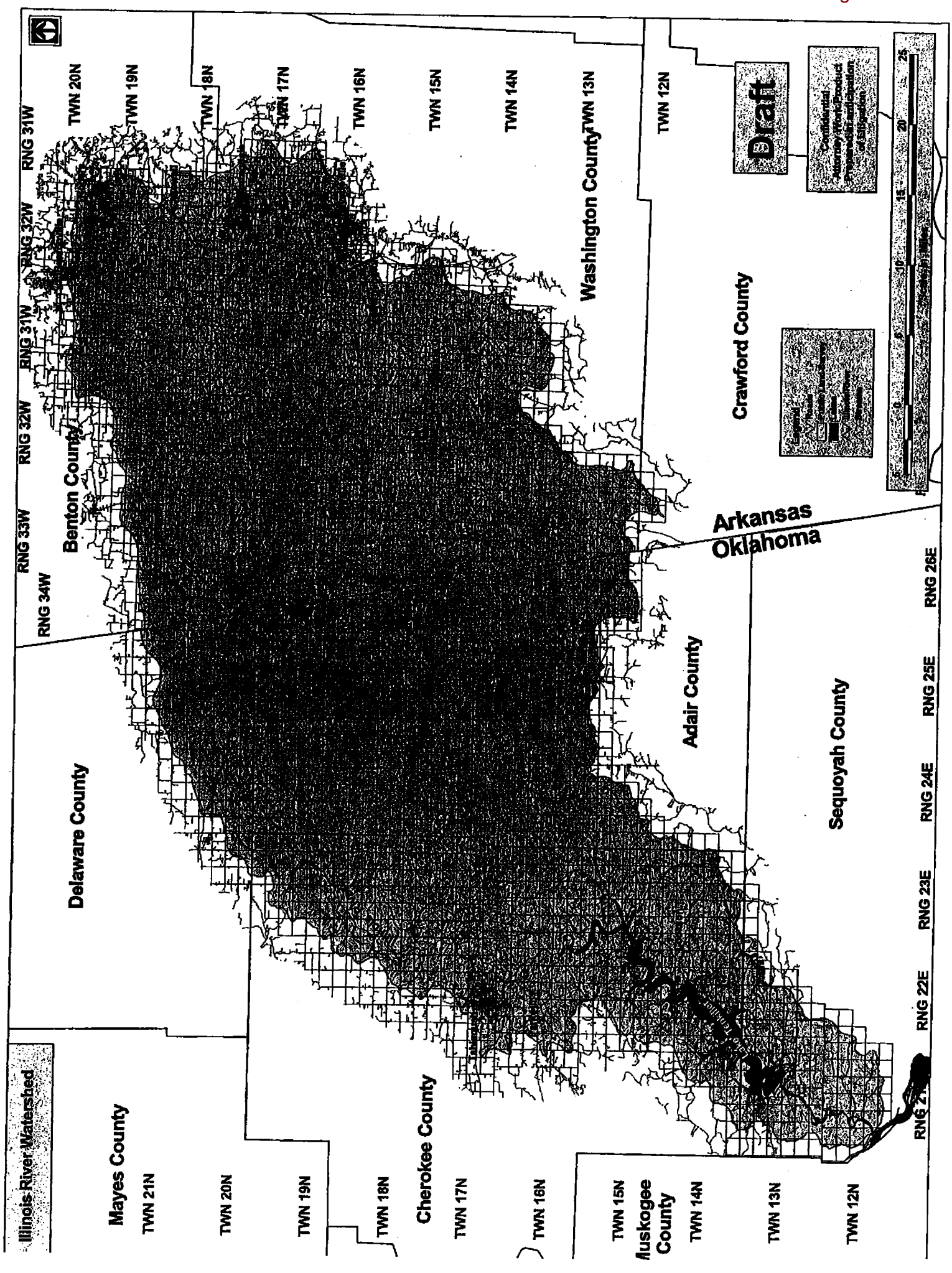

Draft
Confidential
Attorney Work Product
Prepared in anticipation
of Litigation
Arkansas
Oklahoma
Benton County
Washington County
Crawford County
Delaware County
Adair County
Sequoyah County
Cherokee County
Mayes County
Muskogee County
Illinois River Watershed
TWN 21N
TWN 20N
TWN 19N
TWN 18N
TWN 17N
TWN 16N
TWN 15N
TWN 14N
TWN 13N
TWN 12N
RNG 34W
RNG 33W
RNG 32W
RNG 31W
RNG 22E
RNG 23E
RNG 24E
RNG 25E
RNG 26E

Exhibit B

Chain of Custody Record

Miller & Keffer
222 South Kenosha Ave.
Tulsa, OK 74120

Illinois River Watershed
Soil and Litter Manure Sampling

No. 0001

Send to: CDM
2714 Walnut Street
Denver, CO 80205

Sample Placed in Cooler/Bag	Sample ID	Sample Date	Sample Time	Sample Matrix (S=Soil; W=Water; L=Litter)	Sample Type (G=Grab; C=Composite)	Analysis Request*	Comments	Sample Received by Lab
☐	LAL1-A-1-2			Soil	G	Compositing		☐
☐	LAL1-A-2-2			Soil	G	Compositing		☐
☐	LAL1-A-3-2			Soil	G	Compositing		☐
☐	LAL1-A-4-2			Soil	G	Compositing		☐
☐	LAL1-A-5-2			Soil	G	Compositing		☐
☐	LAL1-A-6-2			Soil	G	Compositing		☐
☐	LAL1-A-7-2			Soil	G	Compositing		☐
☐	LAL1-A-8-2			Soil	G	Compositing		☐
☐	LAL1-A-9-2			Soil	G	Compositing		☐
☐	LAL1-A-10-2			Soil	G	Compositing		☐
☐	LAL1-A-11-2			Soil	G	Compositing		☐

Total Number of Samples: 20

END OF SUBMITTAL

Additional Comments: _____

_____

_____ _____ _____ _____ _____

Relinquished by (Signature and Company) **Date/Time** **Received by (Signature and Company)** **Date/Time** **Sample Condition upon Receipt**

_____ _____ _____ _____ _____

Relinquished by (Signature and Company) **Date/Time** **Received by (Signature and Company)** **Date/Time** **Sample Condition upon Receipt**

April 19, 2005

Copies: Pink - Retained by Sample Coordinator; Yellow - Retained by laboratory; White - Included in analytical report

Chain of Custody Record

Miller & Keffer
222 South Kenosha Ave.
Tulsa, OK 74120

Illinois River Watershed
Soil and Litter Manure Sampling

No. 0001

Send to: CDM
2714 Walnut Street
Denver, CO 80205

Sample Placed in Cooler/Bag	Sample ID	Sample Date	Sample Time	Sample Matrix (S=Soil; W=Water; L=Litter)	Sample Type (G=Grab; C=Composite)	Analysis Request*	Comments	Sample Received by Lab
☐	LAL1-A-12-2			Soil	G	Compositing		☐
☐	LAL1-A-13-2			Soil	G	Compositing		☐
☐	LAL1-A-14-2			Soil	G	Compositing		☐
☐	LAL1-A-15-2			Soil	G	Compositing		☐
☐	LAL1-A-16-2			Soil	G	Compositing		☐
☐	LAL1-A-17-2			Soil	G	Compositing		☐
☐	LAL1-A-18-2			Soil	G	Compositing		☐
☐	LAL1-A-19-2			Soil	G	Compositing		☐
☐	LAL1-A-20-2			Soil	G	Compositing		☐

Total Number of Samples: 20

END OF SUBMITTAL

Additional Comments: _____

Relinquished by (Signature and Company) **Date/Time** **Received by (Signature and Company)** **Date/Time** **Sample Condition upon Receipt**

Relinquished by (Signature and Company) **Date/Time** **Received by (Signature and Company)** **Date/Time** **Sample Condition upon Receipt**

April 19, 2005

Copies: Pink - Retained by Sample Coordinator; Yellow - Retained by laboratory; White - Included in analytical report.

Exhibit C

Sample Location Protocol for Each of the Sampling Areas, A thru D

1. On the map/aerial photo for the facility, divide each LAL or CL into four relatively homogeneous areas of approximately the same size and configuration. See Figure 1. These areas should cover the area where litter has been applied. Rectangular or square shapes over the areas where litter has been applied are the best. However, the designated outlines/boundaries of the area should cover most of the area where litter has been applied. These units are identified as the "Sampling Areas" and designated as "A", "B", "C" and "D". For example at LAL5, the first sampling area will be LAL5-A. Each of the four areas should be a minimum of approximately one acre and a maximum of approximately 10 acres. If the LAL or CL is less than 4 acres in size, consider selecting another LAL or divide the field into only three sampling areas.

2. For each sampling area, pace off the length (L) and width (W). See Figure 2. This can also be measured on the aerial photo. The length will be the longer dimension. The L and W can also be the same (square). If the shape is not a rectangle or square, the L and W should be measured at the average dimensions.

3. A grid pattern of twenty sampling locations will be established on each sampling area, A thru D. Divide W by 8 (W/8). The origin of the grid will be located at a distance of W/8 x W/8 from one corner. See Figure 2. That is, all the sampling points will be insert from the boundary of the sampling area (the insert will be a distance of W/8 on all four sides of the sampling area). Determine the remaining width (RW) by calculating W - W/4 = RW. Divide RW by 3 (RW/3). Determine the remaining length (RL) by calculating L - W/4 = RL. Divide RL by 4 (RL/4). The grid pattern will have dimensions of RW/3 x RL/4. See Figure 2.

   Note: If the application area is contained within a larger field or in one area of a field that is not immediately adjacent to another sampling area, creation of the insert (W/8) around the area is not necessary. This results in simpler calculations.

4. Record the values of L, W, W/8, RW, RW/3, RL and RL/4 on the aerial photo and/or in the log book. When recording in the log book, also make a sketch of the field.

5. Twenty sampling locations on a grid pattern will be located in each sampling area. Locate the origin of the grid at W/8 x W/8 paces (or feet) from one corner of the sampling area. In the example from step No. 1 above, this location will be LAL5-A-1. Place a pin flag at this location. Continue walking (or measuring on the aerial photo) a distance of RW/3 paces (or feet) across the width (small dimension) of the sampling area. Stop and place a pin flag at each location. A total of four locations (LAL5-A-1 thru LAL5-A-4) will be determined along this first line across the sampling area. See Figure 2.

6. Turn 90 degrees and pace (or measure on the aerial photo) RL/4 along the length of the sampling area. This location will be LAL5-A-5. Turn 90 degrees and pace (or measure) a distance of RW/3 across the width of the sampling area. Stop and place a pin flag at each location. Another four locations will be determined on this second line across the width of the sampling area (LAL5-A-5 thru LAL5-A-8).

7. Perform step No. 6 another three times for a total of five lines across the sampling area. This will result in 20 locations. For sampling areas that are not rectangular or square, more (larger distance than average width) or less (smaller distance than average width) than four locations

will be determined on each line across the width of the sampling area. Twenty locations should still be located; however, 18 or 19 locations are still acceptable.

Note: Sampling points may be relocated to the nearest point away from standing water, overly wet soils, machinery, trees or locations with excessive vegetation (any of which would preclude collection of a representative or uncompromised sample). If a sample point is relocated, return to the original point to continue the sampling protocol.

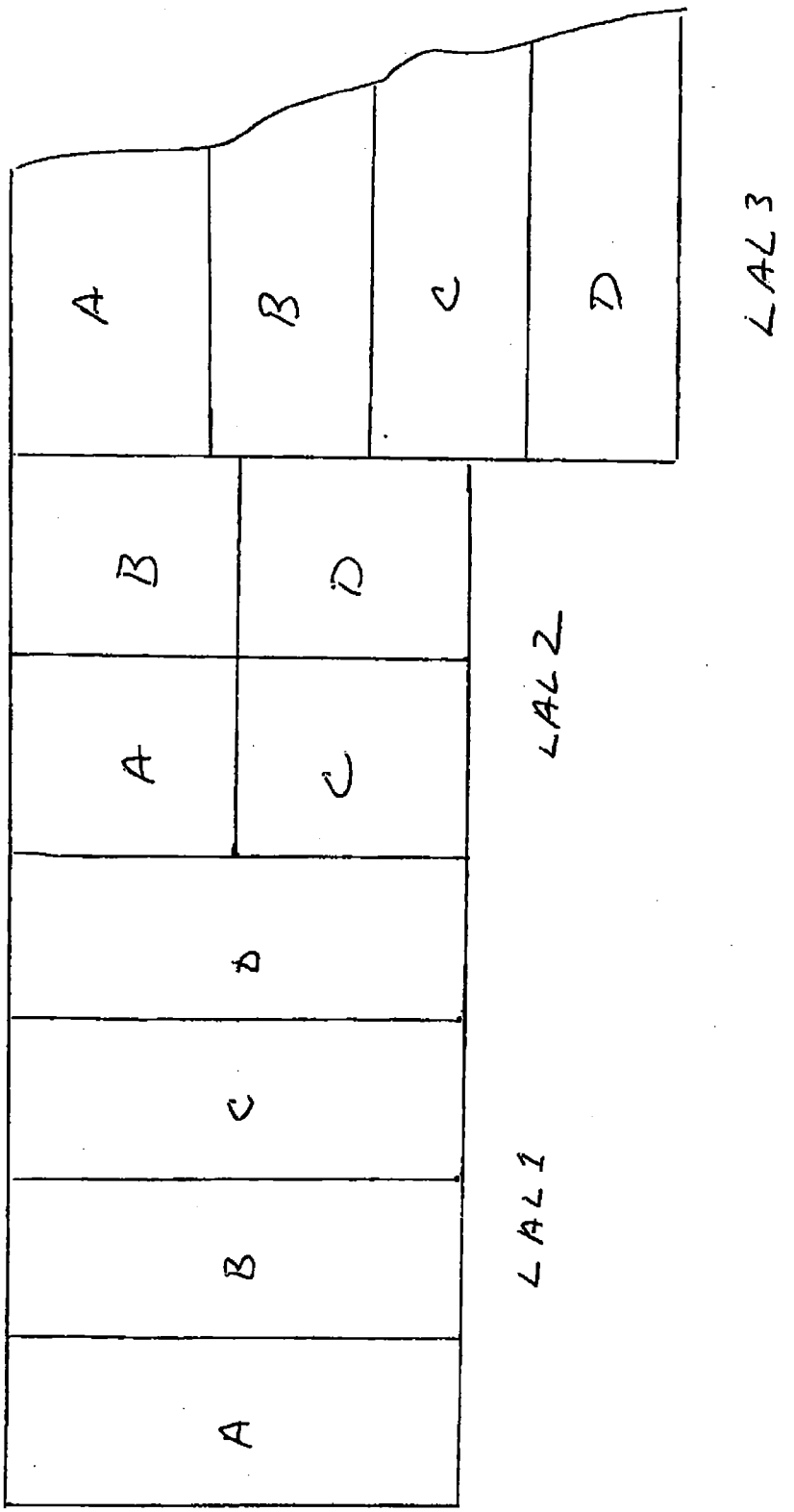


Figure 1

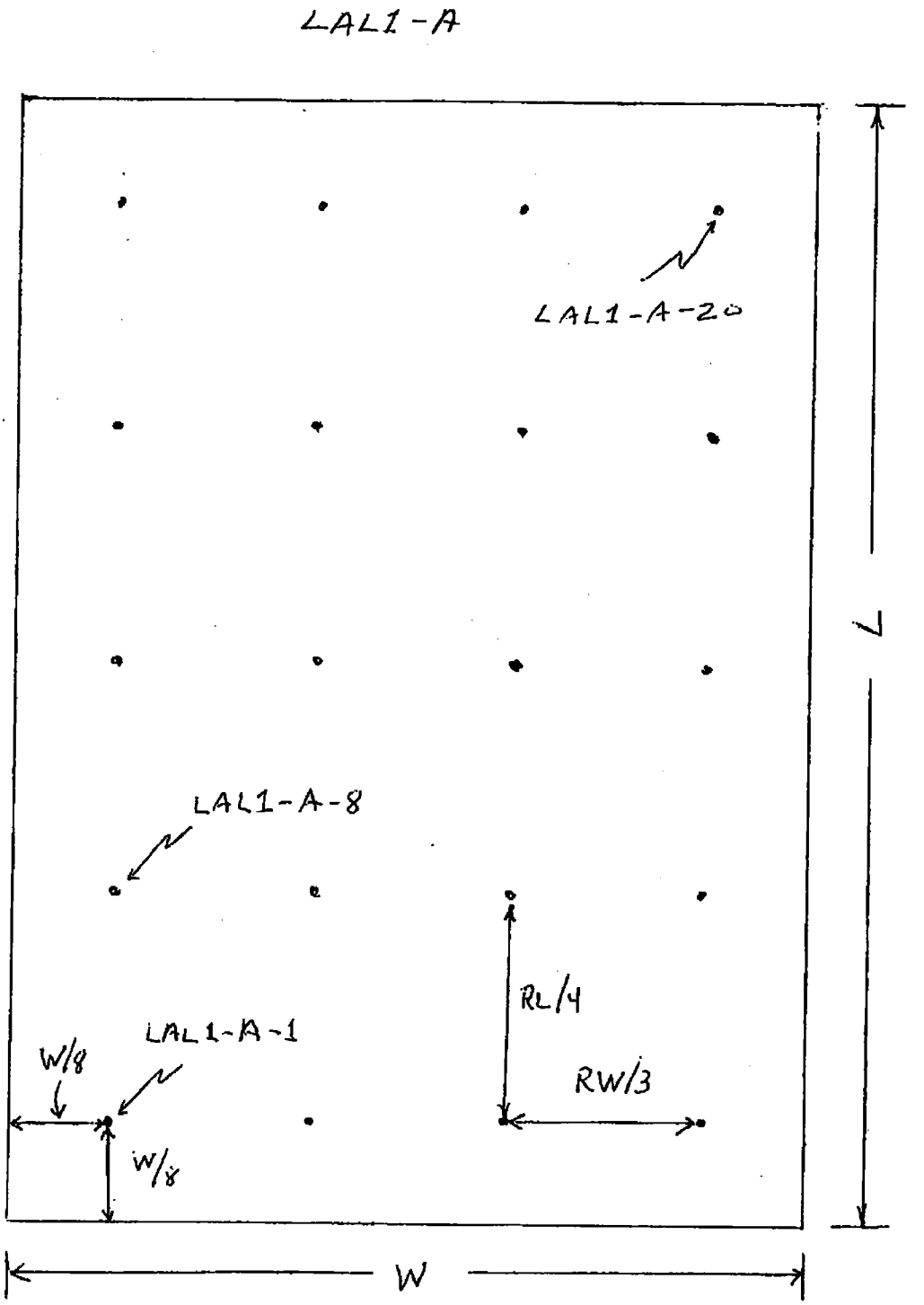


Figure 2

Exhibit D

Illinois River Watershed Soil and Litter Sample Compositing Protocol

1.0 Handling and Compositing of Soil and Litter Samples

All individual soil samples from each sub-location will be placed in individual plastic bags (double bagged), packed in a cooler with blue ice and shipped over night under chain-of-custody to the CDM processing laboratory in Denver, Colorado. The sample number will be located between the inner and outer plastic bag. Each sampling area (up to four sampling areas per LAL) will have 60 individual samples (20 sub-sample locations x 3 sample depths = 60 samples). All samples will be received by the CDM processing laboratory for compositing. Each of the 20 sub-samples will be composited into one homogeneous sample using the protocol described below.

Litter samples will be received by the CDM processing laboratory under chain-of custody in a 5-gallon bucket. The litter sample will be contained in a plastic bag inside of the 5-gallon bucket will be closed with a tie. A unique sample number will be written on the outside of the bucket.

Upon receipt of the samples, the cooler/bucket temperature will be measured using a NIST traceable thermometer. The samples soil will then be removed from the cooler and checked against the chain-of-custody to ensure that all samples have been received.

The twenty sub-samples associated with the individual sample depths or the entire litter sample will be poured into a stainless steel bowl or 2.5-gallon bucket ready for mixing. All equipment will be decontaminated/sterilized with laboratory grade distilled water and 10 percent bleach (see procedure below).

1.1 Mixing of Soil Samples

- All health and safety protocol will be followed as described in the Health and Safety Plan for the Illinois River Basin Project. This is includes wearing nitrile gloves and processing soil in the hood.
- All feathers, rocks, twigs, debris and vegetation will be removed before sieving and mixing.
- Mixing will be accomplished using a disposable, plastic sampling scoop or a decontaminated stainless steel spoon.
- All clods over 0.5 inches in diameter will be disaggregated into smaller particles by hand or the use of a decontaminated stainless steel spoon or mortar.
- If the moisture content is too high to allow homogenization or disaggregation of the particles, the sample will be placed in steel

drying pan and air dried over night.

- The sample will be hand mixed using the plastic scoop or stainless steel spoon for at least five minutes or until particles are uniform in size.
- If a plastic bucket is used, the bucket will then be sealed and inverted or rotated at least 10 times.
- After mixing, the sample will be sieved to remove particles sizes of greater than 2 mm using a decontaminated US Sieve no. 10 (gravel size particles will be removed).
- Each fraction (greater than 2 mm and less than 2 mm) will be weighted. The less than 2 mm fraction will be placed in a plate grinder and reduced in size to 0.074 mm (US sieve no. 200, very fine sand).
- The ground sample will be split using a riffle splitter and sent to the various laboratories (see splitting procedure in section 1.3.1, Duplicate Samples).

1.2 Mixing of Litter Samples

The same procedure as described above for the soil will be used for the litter. However, grinding may not be necessary if the litter can be sieved directly through a US sieve no. 200.

1.3 Laboratory QA/QC Samples (Soil)

Laboratory QA/QC samples will consist of duplicate samples, blind standards, and decontamination rinsate samples. The following describes each type of QA/QC sample.

1.3.1 Duplicate Samples (created at the soil processing lab)

After sample mixing, sieving and grinding, two split samples will be collected. The sub-sample splits should be collected using a nonbiased riffle splitter. The sample is poured through the riffle splitter and into the decontaminated collection pans. The amount of soil or litter contained by the sample container shall be sufficient for the chemical and physical analyses to be conducted.

At least one set of duplicate soil samples will be created for each LAL sampled or at a frequency of one duplicate for every 10 samples sent to the laboratory (which every frequency is less).

At least one set of duplicate soil samples shall be submitted for each CL sampled or at a frequency of one duplicate for every 10 samples sent to the laboratory (which every frequency is less).

A set of duplicate soil samples for a sampling area within a LAL or a CL shall comprise:

- A duplicate sample prepared from the four to six-inch samples.
- A duplicate sample prepared from the two to four-inch samples.
- A duplicate sample prepared from the zero to two-inch samples.

One duplicate sample per integrator will be prepared for the litter samples.

1.3.2 Blind Standards

A blind standard of a certified reference soil shall be sent to the analytical laboratory for every 50 soil samples send to the laboratory. One blind standard will be sent to the analytical lab for every 10 litter samples sent to the laboratory. The blind standard will be sent by the CDM soil processing lab. Blind standards will be for metals, arsenic, and phosphorous.

1.3.3 Decontamination Blanks

A sample of the final decontamination rinsate shall be collected and forwarded to the analytical laboratory for analysis at a frequency of one decontamination rinsate for every 25 samples sent to the laboratory. The decontamination rinsate blank will be generated in the CDM processing laboratory using a final rinse of laboratory grade distilled water. All parameters will be analyzed.

2.0 Shipment of Samples To the Analytical Laboratory

- Once placed in sampling containers (plastic bags or jars), samples shall be held at 4° C on blue ice (sealed in plastic bags) within insulated protective containers.
- If possible, samples shall be shipped immediately via overnight shipment to the analytical laboratory.
- In no event, shall samples be held more than 24 hours before shipment.
- Samples shall be sent to the laboratory under a Chain-of -Custody.
- A custody seal will be place on the outside of the cooler between the lid and the body of the cooler. The custody seal will be signed.
- The Chain-of-Custody shall be sealed in a plastic bag and placed within the insulated protective container holding those samples to which it refers.

3.0 Decontamination of Processing Equipment

All nondisposable equipment (bowls, sieves, spoons, and grinders) will be decontaminated/sterilized after each composite sample is created. Decontamination will include washing with phosphate free water followed by rinsing with laboratory grade distilled water. A final rinse of

10 percent bleach will be performed. The equipment will be air dried.

4.0 List of Analytes and Bottle Requirements

4.1 Analytical Parameters

Soil samples will be analyzed for one of two sets of parameters (short list or long list). Table 1 provides the parameters and analytical methods for the short list and Table 2 provides the parameters and analytical methods for the long list.

Litter samples will be analyzed for Table 2 parameters.

Table 1: Short List Parameters - Soil

Parameter	Method
Moisture content (%)	Gravimetric (105C)
Organic matter	Walkley-Black (Modified)
Soil pH	Water 1:1
Soil Conductivity	Water 1:2
Total Nitrogen	Kjeldahl, modified
Total Aluminum (Al)	EPA SW-3050/6010
Total Phosphorous (P)	EPA SW-3050/6010
Total Arsenic (As)	EPA SW-3050/6010
Total Copper (Cu)	EPA SW-3050/6010
Total Zinc (Zn)	EPA SW-3050/6010

Table 2: Long List Parameters – Manure and Soil

Parameter	Method
Moisture content (%)	Gravimetric (105C)
Organic matter	Walkley-Black (Modified)
Texture (% sand, silt and clay)*	Hydrometer ASTM-D422
Soil pH	Water 1:1
Soil Conductivity	Water 1:2
Total Phosphorous (P)	EPA SW-3050/6010
Mehlich-III Phosphorous (Mehlich-III P)	Mehlich III (ICP)
Soluble Phosphorous	Water 1:10, Bull.396, pg 17
Soluble nitrate	Water 1:10
Total Nitrogen	Kjeldahl, modified
Soluble ammonium	Water 1:10

Soluble sulfate	Water 1:10
Soluble chloride	Water 1:10
TAL Metals	EPA SW-3050/6010
Total Molybdenum (Mo)	EPA SW-3050/6010
Bacteria:	
Total coliform	SM-9221B
enterococcus	SM-9221F
Fecal coliform	SM-9230B
e-coli	SM-9221F
staphylococcus	MPN
campylobacter	MPN
salmonella	MPN
17β-estradiol, estrone, estriol	LC-MS-MS

*split before sieving and grinding

4.2 Anticipated Number of Samples

Soils: Assuming four areas and three depths will be collected at 15 LALs and 8 CLs, 276 soil samples (23 fields x 4 areas x 3 depths) will result (after compositing) and be analyzed. In addition, 28 duplicates (1 in 10); 11 decon rinsates (1 in 24); and 6 standards (1 in 50) will be collected. Two samples for each LAL and CL (23 x 2 = 46) will be selected for Table 2 analyses. Five of the duplicates, one of the standards and 2 of the rinsate samples will be analyzed for Table 2. The remaining samples (230 soil samples, 23 duplicates, five standards and 9 rinsates) will be analyzed for Table 1 parameters. In total, 321 samples will be analyzed (267 for Table 1 and 54 for Table 2 parameters).

Litter: Assuming three houses from each of five integrators, 15 litter samples will be collected. In addition, 5 duplicates, two decon rinsates and two standards will be analyzed. In total, 24 samples will be analyzed for Table 2 parameters.

In addition selected samples will also be analyzed using molecular methods (see task 3.9, Bacterial Analyses by Molecular Methods).

4.3 Bottle Requirements

Soil samples will be analyzed for one of two sets of parameters (short list or long list). Table 3 provides the parameters, bottle requirement and laboratory for the short list and Table 4 provides the parameters, bottle requirement and laboratory for the long list.

Litter samples will be analyzed for Table 4 parameters.

Table 3: Short List Parameters - Soil

Parameter	Bottle	Laboratory
Moisture content (%)	1 quart glass	A&L
Organic matter	1 quart glass	A&L
Soil pH	1 quart glass	A&L
Soil Conductivity	1 quart glass	A&L
Total Nitrogen	1 quart glass	A&L
Total Aluminum (Al)	1 quart glass	A&L
Total Phosphorous (P)	1 quart glass	A&L
Total Arsenic (As)	1 quart glass	A&L
Total Copper (Cu)	1 quart glass	A&L
Total Zinc (Zn)	1 quart glass	A&L

Note: 1 bottle for all of the above analysis

Table 4: Long List Parameters – Manure and Soil

Parameter	Bottle	Laboratory
Moisture content (%)	1 quart glass	A&L
Organic matter	1 quart glass	A&L
Texture (% sand, silt and clay)*	1 quart glass (separate from the other bottles)	A&L
Soil pH	1 quart glass	A&L
Soil Conductivity	1 quart glass	A&L
Total Phosphorous (P)	1 quart glass	A&L
Mehlich-III Phosphorous (Mehlich-III P)	1 quart glass	A&L
Soluble Phosphorous	1 quart glass	A&L
Soluble nitrate	1 quart glass	A&L
Total Nitrogen	1 quart glass	A&L
Soluble ammonium	1 quart glass	A&L
Soluble sulfate	1 quart glass	A&L
Soluble chloride	1 quart glass	A&L
TAL Metals	1 quart glass	A&L
Total Molybdenum (Mo)	1 quart glass	A&L
Bacteria	1 - 250 mL plastic (sterilized)	EML

17β-estradiol, estrone, estriol	1 – 4oz. glass	GTL

*split before sieving and grinding

5.0 Analytical Laboratories

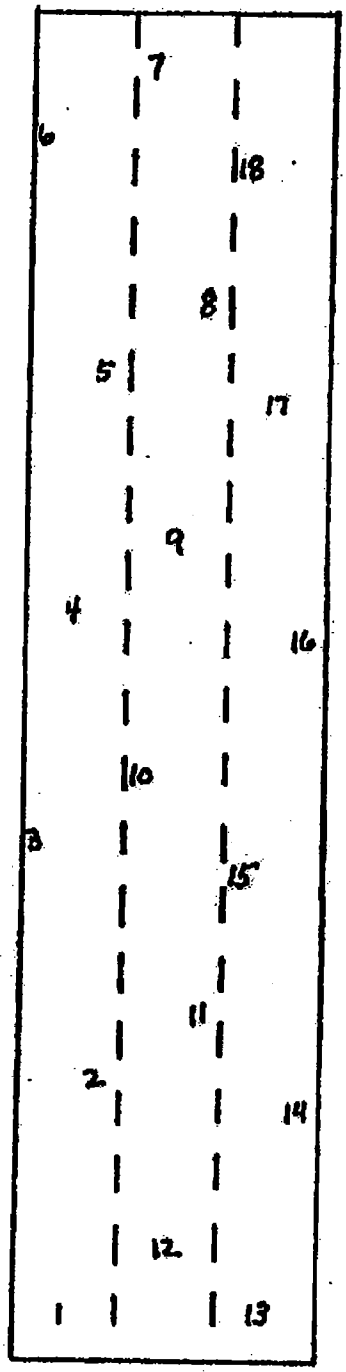
Bottles for estrogen metabolites (all samples) will be shipped to:
General Engineering Laboratories, LLC
741 Corporate Circle, Suite I
Golden, CO 80401
Contact: Paul Winkler, 720-253-3093
Paul.winkler@gel.com

Bottles for nutrients, metals, etc (all samples) will be shipped to:
A&L Analytical Laboratories, Inc.
2790 Whitten Rd.
Memphis, TN 38133
Contact: Scott McKee, 800-264-4522
smckee@allabs.com

Bottles for bacteria analyses from soil and litter will be shipped to:
Environmental Microbiology Laboratory
1150 Bayhill Drive, Suite 100
San Bruno, CA 94066
Contact: Cole Mackelprang, 858-268-2762
cmackelprang@emlab.com
Contact (microbiologist): Mark Wallin, 650-742-8132
mwallin@emlab.com

Bottles for molecular analyses will be shipped to:
North Wind, Inc.
1425 Higham St.
Idaho Falls, ID 83402
Contact: Tamzen W. Macbeth, 208-528-8718
tmacbeth@northwind-inc.com

Exhibit E



TYPICAL LITTER
SAMPLING PATTERN

Steps for Pulling Litter Samples
(see attached house diagram)

1. mentally divide your house into thirds, length wise;
2. take 3 or 4 paces and use sharp shooter to "pull" a small sample (about 1 pint) of litter from "left" 1/3 of the house and place it in the 5 gallon bucket, be sure to obtain litter from the entire depth of the bedding material;
3. take about 20-25 paces and obtain another subsample - place in bucket and mix with other subsample;
4. repeat step 3 until you reach the end of the house - you should end up having pulled about 6 subsamples - 2 each from around the waterers, the feeders and the walls on each side of the house;
5. walk a zigzag pattern down the center of the house pulling and mixing subsamples about every 20-25 paces - you should obtain as many subsamples here as you did in the "left" 1/3 of the house;
6. repeat steps 2, 3 & 4 for the "right" 1/3 of the house;
7. label 1 baggie with the following information:
   your name
   date
   sample type (ex. broiler full house)
   sample number (1, 2 or 3)
8. thoroughly mix the 18 subsamples obtained from the house and place a small amount in the baggie - repeat this process until the baggie is about 1/2 full;
9. repeat steps 2-8 two more times;
10. take samples to your County Extension Agent for mailing to the lab for testing;
11. your results will be mailed to you in about 7-10 days;
12. to obtain your average nutrient value add the 3 values for each of the nutrients (those listed under "lbs/ton on as is basis" at the bottom of the page) and divide by 3;
    about 25% of the nitrogen will be lost after being land applied - to determine the amount of nitrogen remaining in your litter after the losses occur, multiple the average figure by 0.75;
    not all of the nitrogen and phosphorus will become immediately available to your forages - to determine the amount of these nutrients that will become available within about 3 months, divide the remaining nutrients figure by 2;
    all of the potassium (potash) will become available in a matter of days;

F-2248

OKLAHOMA COOPERATIVE EXTENSION SERVICE

Sampling Animal Manure

Hailin Zhang
Waste Nutrient Management Specialist

Douglas W. Hamilton
Waste Management Specialis

James G. Britton
Area Poultry Specialist

Oklahoma Cooperative Extension Fact Sheets are also available on our website at: **http://www.osuextra.com**

The accuracy of a chemical analysis is only as good as the sample sent to the lab. The sample collected should closely represent the material used as a fertilizer. Manure collected at one point in the system may be completely different from manure collected at another point. Manure characteristics can also change with the seasons. Sample and analyze manure close to the time when it will be used. If you only use it during a certain time of the year, sample during that time. Take samples at least once per year and whenever manure handling procedures change. If manure is used throughout the year, sample more frequently. Many laboratories supply sampling kits on request. Always consult with the lab before collecting samples. The representative sample collected may become useless, if the proper shipping and preservation procedure is not used.

Sampling Techniques

Litter Inside a Broiler or Pullet House

Dry litter varies across the width of the house—material near the curtains is different from that under feeders and waterers. There are also differences between brood and growout areas and even the north and south sides of a house. These differences must be considered to get a representative sample. The following techniques allow samples to be taken with birds in the house.

Trench Method

Using a shovel (a narrow spade works well), dig a trench as wide as the shovel across half of the broiler house (Figure 1). Start at the center line of the house and dig a trench in the litter to the sidewall. If there is cake, cut the caked litter to the width of the shovel and collect it too. Place the entire contents of the trench on a tarp or drop cloth. Thoroughly mix the litter using a hoe. Place a portion of this well-mixed litter into a zipper-closing plastic bag. Place it in a second bag. Use the litter remaining on the tarp to backfill the trench.

Zigzag Method

Walk the entire house in a zigzag pattern (Figure 1) and grab 15 to 20 subsamples with a shovel or coffee can. Collect the entire depth of the litter, but be careful not to remove soil beneath the litter. Place subsamples in a plastic bucket, and mix thoroughly. Take a small sample from the bucket and place in a zipper-closing plastic bag. Place in a second plastic bag.

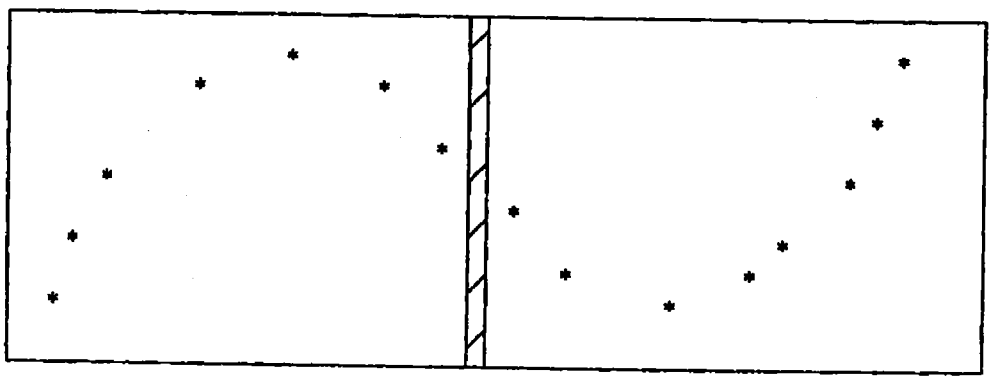


Figure 1. Taking poultry litter samples in the house using trench and zigzag methods.

Litter Inside a Breeder House (partially slatted)

A composite sample from a partially slatted breeder house can be sampled by collecting sub-samples from both slatted and litter areas. In all collect at least 20 sub-samples to get a representative sample of the building. Since 2/3 of the house is under slats, and 1/3 is litter area, collect 14 cores from under the slats and 7 samples from the litter area. Sample through the slats using a soil probe or section of pipe. Collect litter samples similar to the zigzag method above. Place slat and litter samples in a plastic bucket and mix thoroughly. Take a small sample from the bucket and place in a zipper-closing plastic bag. Place in a second plastic bag.

Lagoon Effluent

If you only pump effluent from the top of a lagoon, you only need to take a sample from the upper two feet. Samples taken from the upper layer of the lagoon should represent the contents of the layer for several weeks, although lagoons do change from month to month. It is a good idea to sample lagoon effluent during the season of year you intend to irrigate. For instance, if you plan to irrigate bermudagrass in May and wheat in August, then take two effluent samples—April-May for the bermudagrass, and July-August for the wheat.

Bucket-Toss Method

A simple effluent sampler is a rope attached to a small plastic bucket. Throw the bucket out into the lagoon and let it sink. Slowly pull the bucket back to shore, being careful not to collect scum or solids with the sample. Then swirl the bucket and pour a subsample into a plastic container.

Dipper Method

Dipping is less accurate than the bucket-toss method. But if you object to handling an effluent covered rope, use a plastic bottle securely taped to a long pole. Make sure the pole is long enough to reach over any scum collected at the edge of the lagoon. Dip out a number of samples at different depths and locations, then mix the samples together in a bucket. Swirl the bucket and pour a subsample into a plastic container.

Entire Lagoon Contents

Sometimes, producers need to analyze the entire contents of a lagoon, or they need to measure chemicals deeper than two feet in the lagoon. Lagoons separate into layers (Figure 2). The bottom of the lagoon contains sludge. A scum or crust may form at the top of the lagoon. Between the sludge and scum is a large volume of liquid. To determine the total contents of a lagoon for diagnostic purposes, you must put together a sample from all the layers. You have two choices—collect a complete column of the lagoon profile or collect material from each layer and mix it into a composite sample based on the mass of each layer. Either way means getting out on the lagoon in a boat.

Column Sampler

A number of column samplers are commercially available. All are basically a long hollow tube (Figure 2) that is slowly lowered into the lagoon. Once the sampler reaches the bottom, the tube is closed off, so you can raise the entire column from the lagoon. Be sure the sampler is long enough to reach the bottom of the lagoon and wide enough to collect an undisturbed sludge sample.

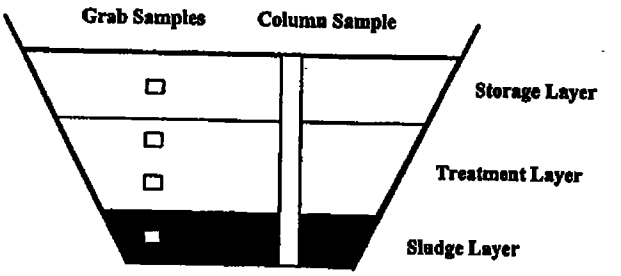


Figure 2. Sampling entire lagoon contents.

Grab Sampler

A discrete or grab sample is a small sample taken from one layer (Figure 2). The idea is to grab the sample without disturbing layers above or below it. Discrete samplers use water pressure to force sludge or liquid into the sampler. The "Sidewinder" sampler is an easy to build grab sampler for lagoons. (Plans are available. Contact your county Extension educator). Once collected, discrete samples may be analyzed separately or combined into a composite sample for the whole lagoon.

Slurry From a Waste Storage Pond or Settling Basin

Layers form in a waste storage pond just as they do in a lagoon. Sampling the entire contents of the pond requires the same techniques as a lagoon. Storage ponds are mixed before slurry is spread on the field as fertilizer. You can use the bucket-toss or dipper methods to collect samples from ponds. But the pond must be agitated first! Solids contents change as the pond is pumped. Take small samples over the entire pumping period and mix into a larger sample. Remove a small subsample from the well mixed sample and place in a plastic container.

Slurry From Pre-fabricated Storage Structures

Above ground storage structures are agitated before spreading. The return line on a pump agitator should have a valve to allow you to take samples. Take a number of small samples while emptying the storage structure. To collect samples from a propeller-agitated pit, use the bucket-throw or dipping method. Remove a small subsample from the well-mixed sample and place in a plastic container.

Slurry or Semi-Solid From Pits Beneath Slotted Floors

Column samplers that are used to sample lagoons work in storage pits as well. Homemade column samplers work just as effectively, though. Take a section of plastic pipe narrow enough to slip through the floor slots, but wide enough to collect undisturbed solids. Lower the pipe through the slots until you feel the bottom of the pit. Cap the upper end, trapping a column of manure. Empty the entire contents of the pipe into a plastic bucket. Take samples from a number of locations

throughout the pit. Swirl or mix the contents of the bucket and pour a subsample into a plastic container.

Solid and Semi-Solid Manure Off Feedlot Surfaces

Using soil probe, take a minimum of 20 cores randomly from the pen surface. Walk the entire area of the pen in a zigzag pattern to make sure you remove cores from all areas. Be careful to remove only manure and not the hardened soil beneath. Collect cores in a plastic bucket and mix them thoroughly. Take a small sample from the bucket and place it in a zipper-closing plastic bag. Place the bag in a second plastic bag. Manure characteristics change with the age of cattle and other management differences, so you should sample representative pens of the same age and similar management practices.

Solid Manure From Stockpiles and Dry Stacks

Using a shovel, remove samples from several locations of at least 18 inches into the pile. Place subsamples in a plastic bucket. Mix, but do not allow the material to dry. Place a portion of the sample in a plastic bag. For added safety, place the bag in a second plastic bag.

Liquid and Slurry During Land Application

Sometimes it is easier to get a representative sample by collecting samples during application. However, the total N concentration of samples collected in the field may be lower than samples taken from storage because some ammonia is lost during application. Contact your local Extension educator or crop consultant before using samples collected in the field for fertilizer recommendations.

Catch Cans in the Field

This technique is especially useful if slurry is spread from a honey wagon or tank truck. Randomly place a number of cans in the field. Collect waste from the cans and mix in a large bucket immediately after spreading. Swirl the bucket to mix the contents and pour a subsample into a plastic container.

Slurry or Liquid From a Big Gun Sprayer

Some big gun sprayers have a valve at the spray riser used to drain the hose. Place a bucket under the valve and open while the gun is running. Open the valve slowly! Big guns operate at high pressures. Collect a number of samples while pumping, and mix together. Take a subsample from the well mixed material and place in a plastic container.

Sample Liquids From a Sprinkler Nozzle

Impact sprinklers and LEPA spray nozzles work at lower pressures than big guns, so it is safe to collect a sample directly from the spray stream. Place a bucket or cylinder directly in the stream. In large irrigation systems, collect a number of samples at different locations. Mix samples into a composite. Take a subsample of the well mixed liquid and place it in a plastic container.

After Collecting Samples

Ship liquid and slurry samples in a quart-sized plastic bottle with a screw top lid. Only fill the bottle half full to allow for gas expansion. Squeeze flexible bottles slightly before screwing on the lid. Place solid and semi-solid samples in zipper-closing plastic bags. Place a second plastic bag over both liquid and solid samples for extra safety. Use cardboard boxes to ship sample bottles and bags. Pack the box tightly with expanded styrofoam peanuts or shredded paper and seal with strapping tape.

Preservatives are generally not needed for manure samples used for fertilizer recommendations. Other analyses may require special shipping and preservation. This is especially true when collecting samples for biological or bacteriological analysis.

F-2207

OKLAHOMA COOPERATIVE EXTENSION SERVICE

How to Get a Good Soil Sample

Hailin Zhang
Director, Soil, Water and Forage Analytical laboratory

Gordon Johnson
Nutrient Management Specialist

Oklahoma Cooperative Extension Fact Sheets are also available on our website at: **http://www.osuextra.com**

Soil tests provide a scientific basis for evaluating available plant nutrients in cropland, pastures, lawns, and gardens. Analyses of soil samples can help farmers and homeowners fine-tune nutrient applications from fertilizers, biosolids, and animal manure. Properly managing the amount of nutrients added to the soil can save money and protect the environment.

Soil nutrients vary by location, slope, soil depth, soil texture, organic matter content, and past management practices, so getting a good soil sample stands out as a major factor affecting the accuracy and usefulness of soil testing. This fact sheet outlines some specific considerations which should be taken into account to get the greatest benefit from soil testing.

Sample Soil at the Right Time

Fields used for production of cultivated crops may be sampled any time after harvest or before planting. Generally, two weeks should be allowed for mailing, analysis, and reporting of results. Additional time may need to be allotted for ordering and application of fertilizers, manure, or lime materials. Noncultivated fields should be sampled during the dormant season. In either case, do not sample immediately after lime, fertilizer, or manure applications because those samples do not represent the true soil fertility.

Fields should be tested annually to measure the available nitrogen pool or as frequently as necessary to gain an understanding of how soil properties may be changing in relation to cultural practices and crop production.

Collect a Representative Sample

Getting a representative sample is simple, but not easy. Research at OSU and other universities has clearly shown that a minimum of 20 cores or small samples taken randomly from the field or area of interest are necessary to obtain a sample which will represent an average of the soil in the field (Figure 1). These cores should be collected in a clean plastic bucket (to avoid metal contamination) and mixed thoroughly by hand. The sample bag should be filled from the mixture. A one pint (OSU soil sample bag full) sample is usually adequate for all tests which might be required. If the sample is too wet to mix, it should be spread out to dry some and then mixed, or sampling should be delayed until the field is drier.

It is important to remember that the sample obtained by the above procedure will be an *average* of the area sampled. If the area sampled is extremely variable in the soil properties which are going to be tested, then it may be better to separate

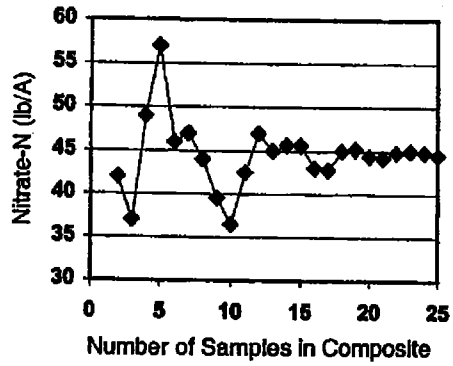


Figure 1. The minimum number of core samples needed to make a representative composite sample is about 20.

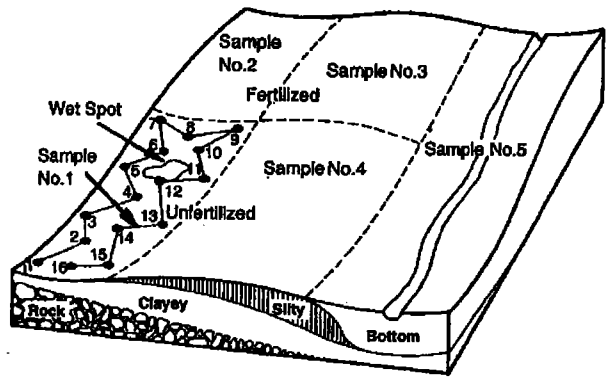


Figure 2. Divide field into uniform sampling areas and follow a random pattern when sampling. Avoid unusual spots and try to obtain a representative sample.

the field into smaller areas, and get a representative (20 cores) sample from each of these areas in order to determine how variable the field is (Figure 2). In this way, it may be possible to treat some areas of the field differently from others and remove variability so that the field can be sampled and treated as a unit in the future. Variability in a field can often be noted by differences in surface soil color and crop growth or yield.

Using only one sample for a large variable field can be very costly. Since the sample represents an average of the soil in that field, recommendations based on the soil test will likely cause the field to be overfertilized on some parts and

underfertilized on other parts. Failure to obtain uniform response to treatments based on a soil test is frequently a result of one sample being used to represent a large variable field.

An example of field variability is shown in Table 1. The range of test values was obtained by testing 40 individual cores taken at random from an "apparently uniform" 80-acre field. The variation is great enough so that for some analyses the average is not a good representation of the field. Areas of the field with the lowest pH, phosphorus, and potassium values will not receive adequate lime or fertilizer if recommendations are based on the average test values.

A single core sample, or spadeful, is extremely risky because it may test anywhere in the range shown for each of the analyses. For example, deficiencies for wheat could range from zero to 37 pounds of P_2O_5 and zero to 34 pounds of K_2O. For alfalfa, which has much greater nutrient requirements, deficiencies could range from zero to 94 pounds of P_2O_5 and zero to 120 pounds of K_2O. This would also affect the amount of nitrogen and lime required. Obviously, unless the 80 acres is divided into less variable units for testing, some areas of the field will receive either too much or too little fertilizer and lime.

In deciding how large an area can be represented by one composite sample (20 cores), the determining factor is not the number of acres involved, but rather, the variability of the area. Some large, uniform fields can be represented well by a single 20-core sample, while some highly variable fields need to be split into two or more smaller areas for testing. Regardless of the field size or main area being sampled, unusual spots in the field (salty or wet spots) should be avoided during the initial random sampling. When unusual spots make up a significant area, they should be sampled separately.

Sample at Proper Depth

Cultivated Fields

For most soil tests the sampling depth is the tillage depth. The reason for this is because most crops have their greatest root activity in the tillage depth. Obtaining a representative sample with regard to depth means that each of the 20 cores taken from an area should be from similar depth, tillage, or six inches. Soil tests are generally calibrated on the basis of an acre furrow slice, approximately two million pounds of soil in the top six inches.

For deep-rooted nonlegumes such as wheat, bermudagrass, sorghum, and cotton, a separate sample representative of the subsoil should be taken in addition to the tillage depth or six-inch sample. This subsoil sample should represent the layer of soil from 7 to 24 inches below the surface. Because nitrate-nitrogen is mobile in the soil, a test of available nitrogen (and/or chloride and sulfate) in the subsoil sample will provide a more complete picture of available mobile nutrients for these crops (Figure 3) and can save fertilizer expenses.

No-till Fields

Noncultivated fields should be sampled to a depth of six inches, again because this is the effective depth of most treatments and the depth of most root activity. Nutrients from fertilizer, animal manure, and lime can be accumulated on the surface if they are surface applied without incorporation. A set of samples from the top two inches will help identify stratification of nutrients and is especially important for pH determination for no-till fields. If nutrient loss in runoff is the main concern, the two-inch sample is better than a six-inch sample because only the surface inch or two is in direct contact with surface runoff.

Salinity Diagnosis

When salt accumulation is suspected as a cause of poor stand establishment and the sample is being taken after planting, then the depth of sampling should approximate the seeding depth (one to three inches). This is especially important when conditions have been favorable for soluble salts to move upward and accumulate near the surface after planting. Since excess salts are most harmful to germination and seedling vigor, it is this shallow depth which should be tested. At other times during the year, a sample of the entire tillage depth may be most useful to test for salt accumulation.

Send Samples for Analysis

Soil sample bags are available at local county Extension offices. Extension offices will mail your samples to the OSU Soil, Water and Forage Analytical Laboratory and assist you to interpret test results.

Table 1. Variability of an 80 Acre Field Based on Soil Tests of 40 Individual Soil Cores .

	Soil Test Values	
Analysis	*Range*	*Average*
pH	4.9-6.3	5.6
Bufferindex	7.1-7.4	7.3
Nitrogen	1-34	11
Phosphorus	23-114	36
Potassium	149-770	306

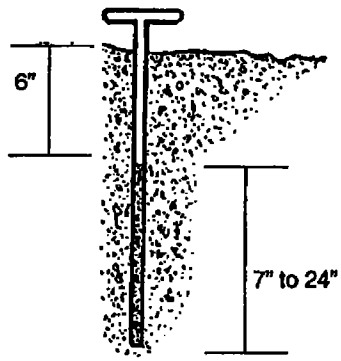


Figure 3. A soil probe is a good tool for obtaining soil samples. Push the tube to the six-inch depth and remove the core. Then take the seven to 24-inch core through the same hole for the subsoil test.

Issued in furtherance of Cooperative Extension work, acts of May 8 and June 30, 1914, in cooperation with the U.S. Department of Agriculture, Samuel E. Curl, Director of Cooperative Extension Service, Oklahoma State University, Stillwater, Oklahoma. This publication is printed and issued by Oklahoma State University as authorized by the Dean of the Division of Agricultural Sciences and Natural Resources and has been prepared and distributed at a cost of 20 cents per copy. 0702

Soil Sampling Protocol

The following steps should be taken to obtain an accurate soil sample for each field.

1. **Obtain the correct soil-sampling tool:** A soil probe marked for a four-inch depth will be used to collect soil samples. A four-inch depth soil core will be collected instead of a six-inch depth core since these soil samples are being used for environmental quality rather than agronomic purposes. If needed, a shovel will be used to collect a uniform soil sample.
2. **Identify sampling area:** Divide field into uniform sampling area based upon the soil types and properties, topography, landscapes, and management history. Select and separate fenced areas, hillsides, creek bottoms, or other well-defined features for accurate and representative soil sampling (Figure 1). Individual pastures make good representative areas if the above uniformity conditions are reasonably met. A soil survey map may be helpful in identifying sampling areas.
3. **Take at least 15 cores (or sub-samples) from each sampling area:** Follow a random zigzag pattern (Figure 1) to collect 15 to 20 individual 4-inch deep sub-samples for an area up to 20 acres and 20 to 25 individual sub-samples for an area greater than 20 acres. Remove thatch and other plant residue from the soil surface before pushing soil probe into the soil. Combine individual sub-samples in a bucket and mix thoroughly to form a sample that represents the entire area. Remove any plant debris and rocks before placing in box. Remove enough of the sample to completely fill the soil sample box (about a pint).
4. **Locations to avoid:** Avoid locations such as old fencerows, low spots, livestock feeding and loafing areas if these areas comprise less than 10 percent of the total sampling area. Such areas may need to be treated as separate sampling areas. If using a shovel, avoid a wedge-shaped soil sample volume.
5. **Handling soil sample:** Record enough information on the soil sample box to identify the location, date of sampling, depth of sample, and test requested. Also record this information in a logbook for future reference. Send soil samples to an approved laboratory for analysis.

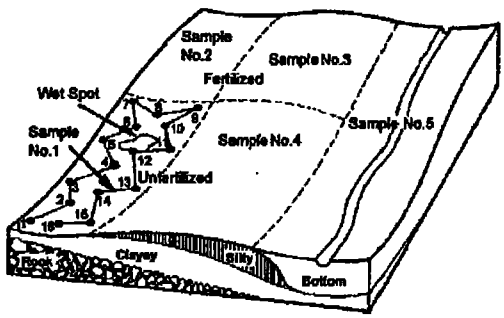


Figure 1. Divide field into uniform sampling areas and follow a random pattern when sampling. Avoid unusual spots and try to obtain a representative sample.

FIELD BOOK PROTOCOL

BEGINNING OF FIELD DAY

Date at the top of the page.
Project Name and/or Site Name
Personnel, their employer, city of origin
i.e. Darren Brown, CDM, Denver, CO
General weather conditions anticipated during the day
i.e. Cloudy, 60's AM, high in 80's. Light winds. Possible rain in PM.
Anticipated activities during this field day
i.e. Anticipated to sample LAL1, LAL2, CL1, and FAC1 today at Farmer John's located in T15N, R23E, Section 12

The above entries must be made at the beginning of each day.

Following Beginning of Day Entries

Time (preferably military i.e. 1620 hours is 4:20 PM)
Field Location (T/R/S) and general location relative to nearest road intersection
i.e. T15N, R23E, Section12, NE of the SE quarter. NW of intersection of Road 2N and 3E.
Owner/operator of property
Maps: Provide two maps - one showing general location of field relative to nearest road intersection and rough distance from intersection. Add any significant landmarks (i.e. house, towers, etc.)

Second map - general shape/dimensions of field and general locations/IDs of subareas to be sampled. General location of sample points.

NOTE: **If you will be providing information on the questionnaire sheets, state this in the field book and identify the Form ID in the field book.**

Activity to be conducted at Site.
i.e. Split field into four subareas and collect samples from 20 locations in each subarea. Sample depths 0-2, 0-4, and 0-6 inches
or Provide operator with 5 gallon bucket and request operator to collect litter samples from 18 locations within the house.

ALWAYS RECORD

Time
GPS Location coordinates (if applicable)
Significant event (i.e. sample collected/sample ID, visitors, change in weather)
Any significant deviations from sampling protocol
Initials at bottom of page
Date at top of page
Page Number

END OF DAY

Sign and date the page.

BLANK PAGES - DO NOT LEAVE BLANK PAGES. SIGN, DATE, AND MARK THROUGH ANY BLANK PAGES.